CLAIMS

What is claimed is:

1. A method of screening for susceptibility to sub-optimal norepinephrine (NE) transport in a subject, the method comprising:

(a) obtaining a biological sample from the subject; and

(b) detecting a polymorphism of a NE transporter gene in a biological sample from the subject, the presence of the polymorphism indicating the susceptibility of the subject to sub-optimal norepinephrine transport.

2. The method of claim 1, wherein the susceptibility of the subject to sub-optimal NE transport is further characterized as susceptibility to orthostatic intolerance.

3. The method of claim 1, wherein the biological sample comprises a nucleic acid sample.

4. The method of claim 3, wherein the polymorphism of the NE transporter polypeptide comprises a G to C transversion within NE transporter exon 9.

5. The method of claim 4, wherein the G to C transversion within exon 9 of the NE transporter gene encodes a NE transporter polypeptide having a proline moiety at amino acid 457.

6. The method of claim 3, wherein the polymorphism is detected by amplifying a target nucleic acid in the nucleic acid sample from the subject using an amplification technique.

7. The method of claim 6, wherein the polymorphism is detected by amplifying a target nucleic acid in the nucleic acid sample from the subject using an oligonucleotide pair, wherein a first oligonucleotide of the pair hybridizes to a first portion of the NE transporter gene, wherein the first portion includes the polymorphism of the NE transporter gene, and wherein the second of the oligonucleotide pair hybridizes to a second portion of the NE transporter gene that is adjacent to the first portion.

8. The method of claim 7, wherein the first portion of the NE transporter gene includes exon 9.

9. The method of claim 7, wherein the first and the second oligonucleotides each further comprise a detectable label, and wherein the label of the first oligonucleotide is distinguishable from the label of the second oligonucleotide.

10. The method of claim 9, wherein said label of said first oligonucleotide is a radiolabel, and wherein said label of said second oligonucleotide is a biotin label.

11. The method of claim 3, wherein the polymorphism is detected by sequencing a target nucleic acid in the nucleic acid sample from the subject.

12. The method of claim 11, wherein the sequencing comprises dideoxy sequencing.

13. The method of claim 3, wherein the step of detecting the polymorphism is detected by contacting a target nucleic acid in the nucleic acid sample from the subject with a reagent that detects the presence of the NE transporter polymorphism and detecting the reagent.

14. The method of claim 13, wherein the reagent detects a G to C transversion within NE transporter exon 9.

15. The method of claim 13, wherein the reagent is an oligonucleotide primer as set forth in SEQ ID NO:9 or SEQ ID NO:10.

16. The method of claim 1, wherein the biological sample comprises a polypeptide sample.

17. The method of claims 1, 2 or 3, wherein the subject is a human subject.

18. An oligonucleotide pair, wherein a first oligonucleotide of the pair hybridizes to a first portion of the NE transporter gene, wherein the first portion includes a polymorphism of the NE transporter gene, and wherein the second of the oligonucleotide pair hybridizes to a second portion of the NE transporter gene that is adjacent to the first portion.

19. The oligonucleotide pair of claim 18, wherein the first portion of the NE transporter gene includes exon 9.

20. The oligonucleotide pair of claim 18, wherein said first and said second oligonucleotides each further comprise a detectable label, and wherein said label of said first oligonucleotide is distinguishable from said label of said second oligonucleotide.

21. The oligonucleotide pair of claim 20, wherein said label of said first oligonucleotide is a radiolabel, and wherein said label of said second oligonucleotide is a biotin label.

22. A set of oligonucleotide primers comprising an anti-sense primer and a sense primer, wherein said oligonucleotide primer set is suitable for

amplifying a portion of the NE transporter gene, wherein the portion includes a polymorphism of the NE transporter gene.

23. The oligonucleotide set of claim 22, wherein the portion of the NE transporter gene includes exon 9.

24. The oligonucleotide set of claim 23, wherein the first portion of the NE transporter gene corresponds to exon 9 of the NE transporter gene.

25. The oligonucleotide primer set of claim 22, wherein said anti-sense primer has a nucleotide sequence selected from the group consisting of SEQ ID NO:33 and SEQ ID NO:34; and wherein said sense primer has a nucleotide sequence of SEQ ID NO:32.

26. A kit for detecting a polymorphism in a gene encoding a NE transporter in a subject, the kit comprising:

(a) a reagent for detecting the presence of a polymorphism of NE transporter gene in a nucleic acid sample from the subject; and

(b) a container for the reagent.

27. The kit of claim 26, wherein the reagent for detecting the presence of the polymorphism of the NE transporter gene comprises a reagent which detects a G to C transversion within NE transporter exon 9.

28. The kit of claim 26, further comprising a reagent for amplifying a nucleic acid molecule containing a polymorphism of NE transporter gene.

29. The kit of claim 28, wherein the amplification reagent include a polymerase enzyme suitable for use in a polymerase chain reaction and a pair of oligonucleotides.

30. The kit of claim 26, wherein a first oligonucleotide of the pair of oligonucleotides hybridizes to a first portion of the NE transporter gene, wherein the first portion includes the polymorphism of the NE transporter gene, and wherein the second of the oligonucleotide pair hybridizes to a second portion of the NE transporter gene that is adjacent to the first portion.

31. The kit of claim 30, wherein the first portion of the NE transporter gene includes exon 9.

32. The kit of claim 26, further comprising a reagent for extracting a nucleic acid sample from a biological sample obtained from a subject.

33. An isolated and purified biologically active human NE transporter polypeptide having an alanine to proline transversion in a transmembrane domain of the polypeptide.

34. The polypeptide of claim 33, further characterized as a recombinant polypeptide.

35. The polypeptide of claim 33, wherein the NE transporter polypeptide comprises an amino acid as essentially set forth in any of SEQ ID NO:2, SEQ ID NO:12 and SEQ ID NO:14.

36. The polypeptide of claim 33, modified to be in detectably labeled form.

37. An isolated and purified antibody capable of preferentially binding to the polypeptide of claim 33.

38. The antibody of claim 37, further characterized as a monoclonal antibody or as a polyclonal antibody.

39. A hybridoma cell line which produces the monoclonal antibody of claim 38.

40. An isolated and purified nucleic acid molecule which encodes a biologically active human NE transporter polypeptide having an alanine to proline transversion in a transmembrane domain of the polypeptide.

41. The nucleic acid molecule of claim 40, further characterized as an isolated and purified cDNA corresponding to exon 9 of a native NE transporter gene and as having a G to C transversion therein.

42. The nucleic acid molecule of claim 41, wherein the encoded NE transporter polypeptide comprises an amino acid sequence set forth in SEQ ID NO:4 or SEQ ID NO:14.

43. The nucleic acid molecule of claim 42, further defined as comprising a NE transporter-encoding nucleic acid sequence as set forth in SEQ ID NO:3 or SEQ ID NO:13.

44. The nucleic acid molecule of claim 43, further characterized as an isolated nucleic acid molecule selected from the group consisting of:

(a) an isolated nucleic acid molecule which hybridizes to a nucleic acid sequence as set forth in SEQ ID NO:3 or SEQ ID NO:13 under wash stringency conditions represented by a wash solution having about 200 mM salt concentration and a wash temperature of at least about 45°C, and which encodes a NE transporter polypeptide; and

(b) an isolated nucleic acid molecule differing from the isolated nucleic acid molecule of (a) above in nucleotide sequence due to

the degeneracy of the genetic code, and which encodes a NE transporter polypeptide encoded by the isolated nucleic acid molecule of (a) above.

45. The nucleic acid molecule of claim 40, further defined as a DNA molecule.

46. The nucleic acid molecule of claim 40, wherein a NE transporter-encoding segment thereof is positioned under the control of a promoter.

47. The nucleic acid molecule of claim 40, further comprising a recombinant vector.

48. The nucleic acid molecule of claim 47, wherein the vector is a recombinant expression vector.

49. A recombinant host cell comprising the nucleic acid molecule of claim 40.

50. The recombinant host cell of claim 49, wherein the host cell is a procaryotic cell or is a eukaryotic cell.

51. A method of preparing a NE transporter polypeptide, comprising: transforming a cell with the nucleic acid molecule of claim 40 to produce a NE transporter polypeptide under conditions suitable for the expression of said polypeptide.

52. A method of detecting in a sample an RNA that encodes the NE transporter polypeptide encoded by the nucleic acid of claim 40, said method comprising the steps of:

(a) contacting said sample under hybridizing conditions with the nucleic acid molecule of claim 40 to form a duplex; and

(b) detecting the presence of said duplex.

53. A method of detecting in a sample a DNA molecule that encodes a NE transporter polypeptide, the method comprising the steps of:

(a) contacting said sample under hybridization conditions with the nucleic acid molecule of claim 40 to form a duplex; and

(b) detecting the duplex.

54. A method of producing an antibody immunoreactive with a NE transporter polypeptide, the method comprising steps of:

(a) transfecting a recombinant host cell with the nucleic acid molecule of claim 40, which encodes a NE transporter polypeptide;

(b) culturing the host cell under conditions sufficient for expression of the polypeptide;

(c) recovering the polypeptide; and

(d) preparing the antibody to the polypeptide.

55. The method of claim 54, wherein the polypeptide comprises an amino acid sequence set forth in SEQ ID NO:4 or SEQ ID NO:14.

56. The method of claim 54, wherein the nucleic acid molecule comprises a nucleic acid sequence set forth in SEQ ID NO:3 or SEQ ID NO:13.

57. An antibody produced by the method of claim 54.

58. A method of detecting a NE transporter polypeptide, the method comprising:

(a) immunoreacting the polypeptide with an antibody prepared according the method of claim 54 to form an antibody-polypeptide conjugate; and

(b) detecting the conjugate.

59. An assay kit for detecting the presence of a NE transporter polypeptide in a biological sample, the kit comprising a first container containing a first antibody capable of immunoreacting with a NE transporter polypeptide of claim 33, wherein the first antibody is present in an amount sufficient to perform at least one assay.

60. The assay kit of claim 59, further comprising a second container containing a second antibody that immunoreacts with the first antibody.

61. The assay kit of claim 59, wherein the first antibody and the second antibody comprise monoclonal antibodies.

62. The assay kit of claim 59, wherein the first antibody is affixed to a solid support.

63. The assay kit of claim 59, wherein the first and second antibodies each comprise an indicator.

64. The assay kit of claim 63, wherein the indicator is a radioactive label or an enzyme.

65. A method for detecting an antibody or fragment thereof, in a sample suspected of containing an antibody or fragment thereof, the method comprising:

(a) contacting the sample with a binding substance comprising a NE transporter polypeptide under conditions favorable to binding of

an antibody or fragment thereof to the binding substance to form a complex therebetween; and

(b) detecting the complex via a label conjugated to the binding substance or via a labeled reagent that specifically binds to the complex subsequent to its formation.

66. The method of claim 65, wherein the binding substance is conjugated with a detectable label and wherein detecting step (b) comprises:

i) separating the complex from unbound labeled binding substance; and

ii) detecting the detectable label which is present in the complex or which is unbound.

67. An assay kit for detecting the presence, in a biological sample, of an antibody immunoreactive with a NE transporter polypeptide, the kit comprising a first container containing a NE transporter polypeptide that immunoreacts with the antibody, with the polypeptide present in an amount sufficient to perform at least one assay.

68. An assay kit for detecting the presence, in biological samples, of a nucleic acid molecule that encodes a NE transporter polypeptide, the kit comprising a first container that contains a nucleic acid molecule identical or complimentary to a molecule of at least ten contiguous nucleotide bases of the nucleic acid molecule of claim 40.

69. A transgenic non-human animal having incorporated into its genome a nucleic acid molecule of claim 40, the nucleic acid molecule being

present in said genome in a copy number effective to confer expression in the animal of a NE transporter polypeptide.

70. The transgenic non-human animal of claim 69, wherein the expression of the NE transporter polypeptide is conferred in cardiac tissue of the animal.

71. A method to enhance transport of NE in a vertebrate subject, the method comprising introducing to a tissue in said vertebrate subject associated with transport of NE a construct comprising a nucleic acid sequence encoding a NE transporter gene product operatively linked to a promoter, wherein production of the NE transporter gene product in the tissue results in enhanced transport of NE.

72. The method of claim 71, wherein the construct further comprises a vector selected from the group consisting of a plasmid vector or a viral vector.

73. The method of claim 71, wherein the construct further comprises a liposome complex.

74. The method of claim 71, wherein the NE transporter gene product comprises a protein having an amino acid sequence as set forth in SEQ ID NO:2 or SEQ ID NO:12.

75. The method of claim 71, wherein the nucleic acid sequence is selected from the group consisting of:

(a) a DNA acid sequence as set forth in SEQ ID NO:1 or SEQ ID NO:11 or its complementary strands;

(b) a DNA sequence which hybridizes to a nucleic acid sequence as set forth in SEQ ID NO:1 or SEQ ID NO:11 under wash

stringency conditions represented by a wash solution having about 200 mM salt concentration and a wash temperature of at least about 45°C, and which encodes a NE transporter polypeptide; and

(c) a DNA sequence differing from an isolated nucleic acid molecule of (a) or (b) above due to degeneracy of the genetic code, and which encodes a NE transporter polypeptide encoded by the isolated nucleic acid molecule of (a) or (b) above.

76. A method for detection of impaired norepinephrine (NE) transport function in a vertebrate subject, the method comprising:

(a) performing a diagnostic test on the subject wherein the diagnostic test is associated with evaluation of NE transport in the subject;

(b) comparing data from the test performed in step (a) to reference data from a reference subject known to have deficient NE transport; and

(c) detecting impaired NE transport in the vertebrate subject if the data from the test subject corresponds to the data from the reference subject.

77. The method of claim 76, wherein the diagnostic test associated with evaluation of NE transport is a tyramine administration test.

78. The method of claim 76, wherein the diagnostic test associated with evaluation of NE transport is a NE clearance test.

79. The method of claim 76, wherein the reference subject has an A457P polymorphism in a gene encoding a NE transporter polypeptide.